Biofouling as a vector of marine organisms on the US West Coast: a preliminary evaluation of barges and cruise ships

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Summary

Recent analyses have shown that ship biofouling is historically one of the strongest vectors of non-native species in marine systems. Moreover, several recently established introductions have resulted from vessel biofouling, underscoring the enduring nature of a vector with origins that are centuries old. Despite an increase in research output over the past decade, there remains a lack of direct measurements of the extent and diversity of organisms associated with vessels' hulls (vessel hull sampling), which is hindering our understanding of the vector. This is especially true of commercial shipping, where options for vector management are being sought and additional data will help inform practical policy solutions. This study adds to the commercial ship biofouling literature by characterizing biofouling of several barges and cruise ships that visited ports in California and the US West Coast.

We sampled seven barges and three tugs involved in Pacific coastwise trade, and five cruise ships also engaged in coastwise transits but some with previous ports-of-call elsewhere (e.g. Hawaii). An additional ocean-going (Coast Guard) Cutter was also sampled on dry dock. In a previous study (Davidson et al., 2009b) we sampled containerships using commercial divers and a remotely operated vehicle. On this occasion, barges were chosen as a focal vessel-type because they contrast with containerships in important characteristics related to biofouling; namely speed, port duration and voyage distances. Cruise ships were also sampled for additional vector data from another ship type with overseas and coastal transits but higher speeds and lower port durations than barges. These cruise ship data are preliminary and further assessments of this vessel type are on-going.

Sampling was carried out on dry docks and in-water by SCUBA divers. For each vessel, we used sample collections and photographs to provide measures of species richness and extent (percent cover or abundance) of biofouling across hull and heterogeneous niche areas (propellers, rudders, stern tubes, thrusters, ladder holes etc). The sea chests of two vessels were also sampled in dry dock.

Among all vessels examined, biofouling cover was less than 20% of submerged surfaces of vessels, with the exception of the Cutter which had approximately 80% fouling cover. Species richness in biofouling communities ranged from 0 to 35 species per ship. At least 169 different species (or distinct taxa) were identified and 25 of those identified to species level are non-native to the West Coast. Algae, barnacles, hydroids, bivalves, polychaetes and bryozoans were the most prevalent taxa among vessels, each occurring on at least ten of the sixteen sampled. Bryozoans (30), amphipods (24), and polychaetes (24) were the most speciose taxa. Among the nonnative species recorded, 16 are already established on the West Coast of North America and a further nine are not yet known to occur on the coast.

As with previous studies, we found biofouling was unevenly distributed across vessel submerged surfaces, with heterogeneous 'niche' areas acting as hotspots for organism accumulation. Hull surfaces were generally free of biofouling and cover was generally low (<10% of the surface area covered). In contrast, the *CG* Cutter had extensive biofouling across all hull surfaces (80% cover), which was probably a function of extended lay-up periods for this vessel in warm waters (Hawaii). Barges, which have fairly uniform hulls because they lack running gears, had hotspots of fouling in ladder holes and dock block areas. Cruise ships had considerable accumulations of biofouling at thruster and stabilizer areas as well as recesses behind specialized propellers (called 'azipods'). We recorded almost three times as many species (14 and 62) in the sea chests of two vessels compared to the outer submerged surfaces of the same vessels.

Biofouling on barges and cruise ships tended to have a larger range of richness and cover compared to containerships from a previous study (Davidson et al., 2009b). The numbers of species ranged from 0-20, 0-33 and 6-35 for containerships, barges and cruise ships respectively. Despite the observed range of species, we cannot conclude from this preliminary comparison that there are significant differences in biofouling richness among vessel types. Additional data are needed before more robust comparisons and analyses can be performed.

Characterizing the richness and abundance of biofouling organisms associated with different types of vessels remains an important component of assessing risk from the commercial-vessel biofouling vector. It is increasingly clear that heavily fouled vessels (those with biofouling covering >10% of submerged surfaces) are in the minority among all vessels sampled in the literature, yet these heavily fouled vessels (outliers) are encountered in many studies. Moreover, the inoculation pressure of vessels with biofouling only in niche areas should be evaluated. In some cases, fouling in niche areas, for example on the grates surrounding bow or stern thrusters, results in tens of thousands of organisms being transferred on a regular basis. These areas are of most concern from a management perspective. Policies that promote additional maintenance attention to niche areas are likely to be effective in reducing propagule delivery via ship biofouling.

Introduction

Vessel biofouling is an important historical and contemporary vector of nonindigenous species (NIS) in marine systems. A recent review by Hewitt & Campbell (2009) ranked biofouling number one for vector strength among marine transfer mechanisms of NIS on a global scale. While historical patterns of shipping and maritime trade are important factors in establishing biofouling as the strongest vector, contributing to 55% of over 1700 species introductions worldwide, there are several examples of recent biofouling-mediated introductions which highlight its continued role in marine invasions (Fofonoff et al., 2003; Davidson et al., 2009a; Hewitt et al., 2009).

Although there has been a recent uptick in scientific output regarding ship biofouling (e.g. Coutts & Taylor, 2004; Davidson et al., 2009b; Lee & Chown, 2009; Sylvester & MacIsaac, 2010; Coutts et al., 2010), there remain critical gaps in evaluating patterns and underlying processes of biofouling transfers across a range of vessel scenarios. Several factors influence biofouling transfers and lead to successful introductions, including voyage routes, antifouling paint condition and effectiveness, hull maintenance practices, seasonal variation, vessel speeds, voyage ranges, port durations, and environmental conditions. Because data are sparse and the number of factors (and their combinations) so numerous, it is difficult to predict how propagule delivery via biofouling is associated with different vessel types and circumstances. These gaps are hindering efforts to manage the vector(s) to reduce coastal marine invasions.

For the present study, our aims were to add to the ship biofouling literature by evaluating biofouling on vessels involved in coastwise trade, including vessels arriving to California for which few data exist. We chose to examine barges because of their slower speeds and higher port durations compared to containerships that we have studied previously. We also sampled cruise ships which have coastal itineraries but travel at faster speeds and with shorter port durations compared to barges. Importantly, we also sampled these vessel types because we were able to get access to them and they act as vectors of fouling species on the West Coast. Our goal was to characterize the extent and richness of algae and invertebrates associated with underwater surfaces of each vessel. This provided a preliminary evaluation of the vector activity by these vessel types in California and elsewhere on the Pacific Coast. The study also provided comparative data to determine whether coastwise vessels tend to have higher levels of biofouling accumulation than ocean-going ships because of their shorter voyages. We previously sampled trans-oceanic containerships (n=22) and found biofouling extent to be quite limited, especially on laminar hull surfaces (Davidson et al., 2009b). Similarly, evaluating cruise ships allowed for an assessment of a vessel type with distinct characteristics (port durations, typical speeds, voyage routes) compared to barges and containerships.

Methods

Vessels were sampled on dry docks in San Francisco and Victoria BC and in-water using SCUBA divers in San Francisco Bay, LA/Long Beach and Anacortes, Washington. Sampling was conducted from January 2009 through February 2010. Both types of sampling involved inspections of below-waterline surfaces of vessels. Vessels were chosen haphazardly based on cooperation and permission by barge and cruise ship operators and the availability in vessel schedules of time windows for sampling. In addition to biological sampling, vessel operators provided information about their vessels and their recent operational history; these data were obtained from the California State Lands Commission hull reporting forms (specifically C. Scianni, CSLC). These data included previous dry docking date, antifouling paints used, typical speeds and port durations, previous ports and voyage details, lay-up periods, and vessel particulars (dimensions or IMO number).

During dry dock sampling, the ship yards provided a window of time, typically about 45 minutes, for surveys as soon as possible after water drained from the dock floor. Photographs, photo-quadrats and samples were taken before power washing began, although in some cases ship yard staff worked concurrently with sampling crews. Sampling had to be tailored to the specific docking situation on the day (e.g. based on height above the ground of hull locations and areas of restricted access), but included evaluations of hull surfaces and niche areas (base of rudder, propeller, thrusters, intake grates). Where possible, photo-quadrats were taken on hull surfaces while photographs were taken of propellers, rudders, stern tubes, thrusters, dock block areas and any other accessible heterogeneous location. For each vessel, a combination of notes, wide angle images (bow area, mid ship, stern area), and haphazard photo-quadrats (16cm x 22cm) of areas of fouling were used to estimate per cent cover of fouling per ship. For two vessels, an opportunity to photograph and collect samples from sea chests (two per ship) was afforded us by ship yard staff who removed the intake-gratings.

In-water sampling of barges and tugs followed a similar protocol and also included photographs, photo-quadrats and sample collections from hull surfaces and niche areas. Diving was carried out from a dive boat and included underwater transects (three divers in a belt transect from bow to stern) and niche area surveys. Tugs were only available for sampling when tug operators granted permission and turned off all running gears. On some occasions during in-water sampling, photo-quadrats were not possible because of limited visibility.

Samples were collected using paint scrapers at each hull and niche area where biofouling was encountered and initially stored in pre-labeled zip-lock bags. For hull surfaces, samples were collected at each hull location where biofouling occurred or across different hull locations (e.g. randomly at bow, midship and stern) if biofouling coverage was extensive across large areas of the hull. Collections aimed to sample the full diversity of organisms encountered visible at each vessel location. Initial examinations of samples were conducted in the laboratory immediately after collection and they were preserved in >70% ethanol, except for some soft-bodied forms (e.g. athecate hydroids and polychaetes) that were preserved in formalin.

The outcome of sampling from dry dock and diver surveys was an estimate of species richness and biofouling extent per vessel with data on distribution of organisms across hull and niche areas. Three different methods of estimating biofouling extent were used:

- A coarse estimation of biofouling percent cover for each vessel was made using *in situ* assessments and photographs of vessels' submerged surfaces.
- A quantitative point-count estimate of biofouling percent cover was made on a subset of vessels (eight) using photo-quadrats (n=10 photo-quadrats per vessel). The replicate quadrats (22cm x 16cm) were taken randomly from mid-ship flat hull surfaces only and excluded niche areas.
- Organism abundance to the nearest order of magnitude (tens, hundreds, thousands, tens of thousands etc) was estimated within niche areas. Samples and photographs were used to determine the numbers of individuals and colonies associated with each niche area examined.

Samples were examined by microscope in the laboratory with each morpho-species assigned to voucher vials and subsequently identified to the lowest taxonomic level possible. Species level identifications of algae were not attempted (these were only differentiated to morpho-species). Identifications of invertebrates were done with the aid of confirmation by several experts. Data were plotted and analyzed to provide comparisons of distributions, richness, extent (cover and abundance) and taxonomic composition among vessels.

Results

Vessel characteristics

There were 16 vessels sampled in total with eight sampled on dry dock and eight in-water using SCUBA. They comprised of seven barges, three tugs, five cruise ships and one Coast Guard Cutter (Table 1). The vessels had a wide range of durations since their most recent dry docking (4 months to 4 years), although nine of the twelve vessels for which these data were available had been out of dry dock for two years or more. Just two vessels reported using a foul-release antifouling paint on portions of their vessel's underwater surfaces while all other vessels reported copper-based antifouling paints (antifouling data were unavailable for four vessels).

The primary voyage pattern reported by all vessels was a Pacific coastwise route (Table 1). Ports located from Mexico to Alaska were listed in itineraries among the 16 vessels. There were three vessels that had overseas (non-coastal) port visits in addition to a general coastwise pattern: *Cruise Ship 5* traveled from the Caribbean through the

Panama Canal prior to its West Coast itinerary while two other cruise ships listed Hawaiian ports as recent stops prior to intra-coastal schedules (Table 1). The longest coastal routes by individual vessels ranged from California to Alaska but the strongest connection, in terms of voyage frequency, was between southern California and San Francisco Bay (primarily by barges). Barge itineraries also provided intermittent links between the port regions of California with the Pacific Northwest. For example, the 22 port visits prior to sampling for *Barge 7* included one transit from Oregon, one from Humboldt Bay, and 18 transits between San Francisco Bay and southern California. Similarly, *Barge 4* reported a recent itinerary consisting of five transits between San Francisco Bay and Southern California, one to Humboldt Bay, and one to Burnaby BC.

There were striking differences among ship types with regard to voyage speeds and port durations. The usual voyage speeds for cruise ships was greater than 14 knots while barges (and tugs) reported less than 11 knots and typically seven or eight knots. Barges and tugs all had typical port durations of greater than 24 hours, two reporting regular port stays of three and four days each. It was also notable that extended idle (lay-up) periods were reported for some barges, including two that anchored in San Francisco Bay for seven and ten days and another (*Barge 6)* that spent 96 days in Coos Bay. Cruise ships, by contrast, typically spent less than 12 hours in port with three days as the longest stationary period reported.

The Coast Guard Cutter reported typical speeds of 12 knots, although this vessel can often attain speeds of >25knots. Exact information on previous ports and activity was not available for this vessel but the opportunity to sample it was taken because of its unusual voyage history (encompassing Hawaii, the South Pacific, and Alaska) and irregular service periods (including extended multiple-week durations in port).

Finally, there were considerable differences in the size and complexity of vessel-types sampled. Barges had an average length of 119m (± 18m) while Cruise ships ranged from 260m to 294m in length (Table 1). Moreover, heterogeneous underwater surfaces or niche areas on cruise ships were numerous and included: three bow and three stern thrusters, mid-ship stabilizers (wings on the starboard and port sides of cruise ships that can extend to offer additional stability and which retract into a recess of several meters length and approximately two meters deep), twin propellers and shafts, rudders and heterogeneous stern areas (with struts). Some cruise ships had 'azipods' or azimuth thrusters instead of regular propellers, eliminating the need for rudders. Barge underwater surfaces are comparatively uniform because they have no running gears, although they have ladder holes and rungs along the sides that can extend below the waterline. Tugs accompanying barges also had very heterogeneous underwater surfaces including complex propeller systems and rows of grates and piping at midship.

Table 1. Vessel characteristics and recent operational histories for 16 vessels sampled on dry dock and in-water.

Vessel	Sampling	Vessel length	Typical Speed (knots)	Typical port duration	Duration since last dry dock	Antifouling	Voyage region (in recent months)
Barge 1	dry dock	101m	7	36-48 hrs	5 years+ *	copper CDP	California - Alaska
Barge 2	dry dock	128m	7	48-96 hrs	4 years	no answer	Puget Sound - Alaska
Barge 3	in-water	115m	8	>24 hrs	3 years	copper CDP	California - Oregon - BC
Barge 4	in-water	115m	8	>24 hrs	2.5 years	copper CDP	California - Washington - BC
Barge 5	in-water	156m	11	>24 hrs	2.5 years	copper CDP	Oregon - Washington - BC
Barge 6	in-water	103m	7	72 hrs	4 months	copper CDP	California - Oregon - Washington
Barge 7	in-water	115m	8	>48 hrs	5 months	copper CDP	California - Oregon
Tug 1	in-water	38m	11	>24 hrs	2.5 years	copper CDP	Oregon - Washington - BC
Tug 2	in-water	39m	8	>24 hrs	no answer	no answer	California - Washington - BC
Tug 3	in-water	35m	8	>24 hrs	no answer	no answer	California - Oregon - Washington
Cruise ship 1	dry dock	292m	17.5	10.5 hrs	3 years	copper CDP	California - Mexico
Cruise ship 2	dry dock	260m	14	8 hrs	3 years	foul release	California - Mexico
Cruise ship 3	dry dock	290m	15	<12 hrs	2 years	foul release & copper SPC	California - Mexico - Hawaii - BC
Cruise ship 4	dry dock	294m	<24	<12 hrs	~ 2.5 years	no answer	California - BC - Alaska
Cruise ship 5	dry dock	260m	15	10 hrs	4 years	copper SPC	Caribbean - Panama - Costa Rica - Mexico - California
Cutter	dry dock	115m	12	variable (3 month on/off cycles)	9 months	copper SPC	Hawaii - Samoa - Alaska - California

* vessel operator indicated that this was the 1st drydocking. The vessel was delivered in 2001.

CDP = controlled depletion polymer

SPC = self-polishing copolymer

Biofouling taxa richness

Macroscopic biofouling organisms were observed and recorded on all vessels except two, which had no detectable biofouling. We did not detect biofouling on *Barge 7* within bow-to-stern transects or niche areas, including ladder holes and dock block areas. This barge had been dry docked five months prior to sampling and had spent time in the interim in riverine (freshwater) ports as well as marine ones. The antifouling paint was unblemished in all locations. Similarly, *Tug 2* (not connected to *Barge 7*) was found to have no biofouling within hull areas and running gear surfaces examined underwater.

The remaining vessels had a diverse range of macro-fouling organisms within biofouling communities (Table 2). One vessel, *Barge 4*, had organisms from nine different phyla within biofouling on its submerged surfaces, while a further seven vessels had six to eight different phyla. Algae occurred on 12 of the 16 vessels with filamentous green algae as the dominant type, although brown and red species were also recorded. The other major taxa, occurring on a majority of vessels, were barnacles, hydroids, bivalves, polychaetes, bryozoans and mobile arthropods (amphipods, decapods, pycnogonids). Ascidians, sponges, flatworms, nemerteans and nematodes each occurred on four or fewer vessels.

Table 2. The presence and absence of taxonomic groups in biofouling of 16 vessels. Presence is denoted by X. Additional data from sea chests of two vessels are also presented (far right columns). The 15 taxonomic groups listed are from 11 different phyla with arthropods, cnidarians, and mollusks divided into two or more sub-taxa.

Taxonomic groups	Barges							Tugs			Cruise ships						Sea chests	
	1	2	3	4	5	6	7	1	2	3	1	2	3	4	5	cutter	barge 2	cruise 3
algae	X	X	X	X	X	X	•	X	•	X	X	X	X	X	•	X	•	•
barnacles	X	X	X	X	X	X	•	X	•	X	X	X	X	X	X	X	X	X
mobile arthropods	X	X	X	X	•	X	•	•	•	•	X	X	X	X	X	X	X	X
hydroids	X	•	X	X	•	X	•	•	•	•	X	X	X	X	X	X	X	X
anemones	X	•	•	•	•	•	•	•	•	•	•	X	•	•	•	•	•	X
bivalves	X	X	X	X	•	•	•	•	•	•	X	X	X	X	X	X	X	X
gastropods	X	•	•	•	X	•	•	X	•	•	X	X	•	•	•	•	•	X
polychaetes	X	•	X	X	•	X	•	•	•	•	X	X	X	X	•	X	X	X
bryozoans	X	•	X	X	•	X	•	•	•	•	X	X	X	X	•	X	X	X
ascidians	•	•	•	X	•	•	•	•	•	•	•	•	•	•	•	•	•	X
flatworms	•	•	X	X	•	•	•	•	•	•	•	•	•	•	•	•	•	•
nemerteans	X	•	X	X	•	•	•	•	•	•	•	•	•	•	X	•	•	•
nematodes	X	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
sponges	•	•	•	•	•	•	•	•	•	•	•	X	•	•	•	•	•	•
insects	X	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	X	X

Species richness ranged from zero to 35 per vessel (Fig. 1a). The two vessels with the highest richness, *Barge 1* with 33 species and *Cruise ship 4* with 35, were sampled on dry dock and had durations of over 30 months since previous dry docking. The taxa responsible for contributing such relatively high richness differed for each vessel; 14 of the 33 species on *Barge 1* were amphipods (8) or polychaetes (6) whereas 18 species on *Cruise ship 4* were barnacles (10) or bivalves (8). Two barges sampled in-water had 31 and 32 species each (*Barge 3* and *Barge 4*, respectively). A further six vessels had between 10 and 21 species on their external submerged surfaces and even distribution of species among taxa (i.e. no taxon dominated). Vessels that had comparatively low species richness (fewer than six species) tended to have isolated patches of barnacles and algae and very little biofouling of niche areas. There was no significant difference in richness between barges and cruise ships (Mann-Whitney U test, test statistic = 40.5, $p > 0.05$). There was no significant difference in richness between vessels sampled on dry dock versus in-water (Mann-Whitney U test, test statistic = 86.5, $p > 0.05$). We also found no relationships between species richness on vessels and duration-since-dry-docking (plot shown in Appendix A).

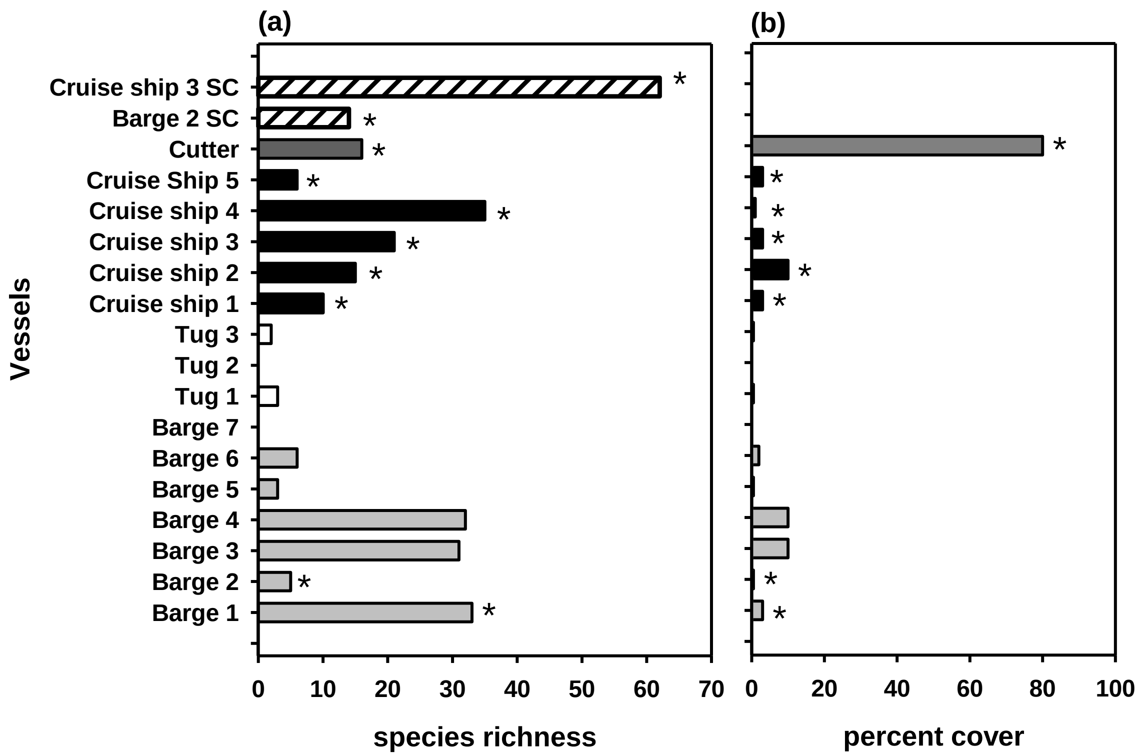

Figure 1. Species richness and percent cover of biofouling from 16 vessels. Species richness (A) and percent cover (B) is plotted for barges (light grey), tugs (white), cruise ships (black), a Coast Guard Cutter (dark grey), and the sea chests of two vessels (stripes; richness only). An asterisk indicates the vessels that were sampled on dry docks. Richness estimates were made from sample collections at each accessible location where fouling was present. Extent (percent cover) estimates were made using coarse evaluations of macro-fouling coverage *in situ* and from images. All organisms have yet to be identified to species level, but distinct morpho-species have been enumerated and vouchered for each vessel.

We recorded 14 and 62 species in the sea chests of *Barge 2* and *Cruise Ship 3*, respectively (Fig. 1a). For both vessels, almost three times as many species were recorded from sea chests as from external surfaces. For *Barge 2*, all of the species that were recorded in external hull biofouling, except filamentous green algae, were also recorded within the sea chests, with amphipods contributing most to the richness of sea chest samples (*Corophium spinicorne*, *Eogammarus confervicolus*, *Gnorimosphaeroma oregonense*, *Corophium* sp1, and *Gammaridae* sp.1). For *Cruise ship 3*, just half of the species found on outer hull surfaces were also found in sea chests. The most species rich taxa within sea chests were 15 polychaete species, 13 bivalves, and six species each of barnacles, amphipods and bryozoans. It was also notable that 11 of the 21 species found on the outer submerged surfaces of *Cruise ship 3* were barnacles.

Among all vessels combined, 169 species or distinct organisms were identified (Appendix B). The richest taxa were bryozoans (30), polychaetes (24), amphipods (24), barnacles (20), and bivalves (19). There were 25 non-native species recorded, including 16 that are already established on some parts of the West Coast and nine that are not known as established NIS on the coast (Appendix B). Seven of the NIS recorded were barnacles but only one, *Amphibalanus amphitrite*, is already established on the West Coast. The other six would represent novel introductions should they become established. Other NIS included *Ciona intestinalis, Botryllus schlosseri,* and *Botrylloides violaceous* (ascidians)*, Monocorophium ascherusicum* and *Elasmopus rapax* (amphipods), *Watersipora subtorquata* and *Bugula neritina* (bryozoans), and the oysters *Crassostrea gigas* and *Ostrea edulis*.

Biofouling extent, abundance and distribution

The percent cover of biofouling on vessels was low (<10%) for a majority of vessels (Fig. 1b). The major exception was the Cutter, which had an even distribution of biofouling algae and invertebrates across hull surfaces (Fig. 2). Although this vessel had been out-of-water for hull maintenance less than a year prior to sampling, its frequent periods of inactivity in Hawaiian waters probably contributed to the extent of biofouling observed.

Figure 2. Biofouling on a Coast Guard Cutter vessel. This vessel was the only one among 16 sampled to have a wide coverage of biofouling across a majority of its hull surface area. From left to right: serpulid and spirorbid tubeworms on hull surfaces; several taxa including tunicates on an intake-grating; tubeworms and bryozoan colonies on hull surfaces.

Point count analysis of biofouling using photo-quadrats underscored the lack of macro-fouling cover on hull surfaces. Bare space and biofilm dominated seven of the eight vessels examined, but the Cutter had substantially greater cover of macro-fouling organisms compared to bare space and biofilm (Fig. 3). Only two other vessels with photo-quadrat data, *Barge 6* and *Cruise ship 3*, had less than 90% bare space, primarily due to filamentous green algal cover.

Although percent cover of organisms on all other vessels was less than 10%, this did not necessarily result in low organism abundance or species richness. The two vessels with the highest species richness had less than three percent biofouling cover of overall wetted surface area (*Barge 1* and *Cruise ship 4*, Fig. 1). The distribution of organisms in niche areas accounted for this pattern. Niche areas typically account for <2% of the submerged areas of vessels but organisms were concentrated around niche areas, sometimes in very high numbers and diverse assemblages.

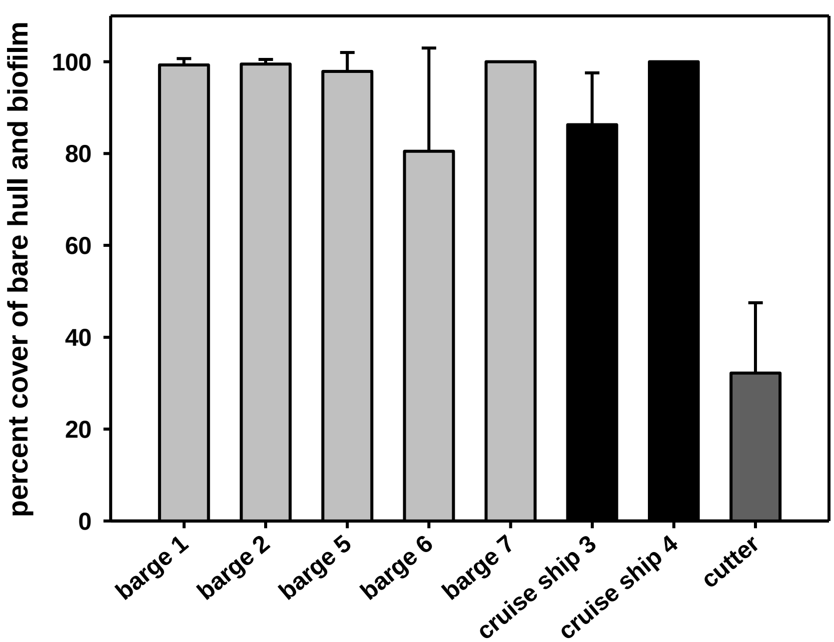

Figure 3. Average percent cover of bare hull and biofilm on hull surfaces of eight vessels. The mean percent and 95% confidence intervals are shown for barges (light grey), cruise ships (black) and a Cutter (dark grey). These areas without macro-fouling organisms were calculated using point counts from 10 random quadrats of flat hull surfaces, excluding all 'niche' areas of each vessel. Only the Cutter had a majority of its hull surfaces covered in macro- algae and invertebrates.

For example, tens of thousands of individual organisms were concentrated around the bow thruster areas, especially the thruster gratings, of four cruise ships (Table 3). Dense accumulations of barnacles (sometimes >500 per $100cm^2$) dominated the biomass of these niche areas and provided a biofouling matrix that supported additional species and secondary colonization. Similar abundances of organisms, also dominated by barnacles, were recorded at the stabilizer areas of *Cruise ship 2*. The azipod backing plate of *Cruise ship 4*, which was removed and placed on the dock floor during sampling, had tens of thousands of individuals dominated by tubeworms and an overall sampled richness of 17 species. As far as we are aware, this type of niche area has not been sampled for biofouling before and is inaccessible to in-water sampling.

Table 3. Biofouling abundance associated with selected heterogeneous niche areas of vessels. The abundance of organisms was estimated in orders of magnitude within niche areas of vessels. Richness and dominant taxa are also listed. Several fouled niche areas among all vessels sampled were not accessible (e.g. out-of-reach on dry dock). Note: examples of unfouled niche areas were also encountered among vessels, including ladder holes of barges, dock block areas, and propellers.

Vessel	Location (niche area)	Estimated abundance (log scale)	Richness	Dominant taxa
Barge 1	ladder hole	10 ^ 3	22	mussels and barnacles
Barge 1	stern struts (notch for tug)	10 ^ 3	24	barnacles and mussels
Barge 2	sea chest	6 x 10^2	14	mussels
Barge 4	ladder hole	10 ^ 3	6	barnacles
Barge 6	dock block areas	10 ^ 3 per block	5	hydroid
Cruise ship 1	thruster grating	10 ^ 4	6	barnacles
Cruise ship 2	bow thruster	10 ^ 4	10	barnacles and mussels
Cruise ship 2	stabilizer	10 ^ 4	n/a	barnacles
Cruise ship 3	sea chest	10 ^ 3	62	barnacles and mussels
Cruise ship 3	bow thruster	10 ^ 4	20	barnacles
Cruise ship 4	azipod backing	10 ^ 4	17	tubeworms
Cruise ship 4	bow thruster	10 ^ 3	21	barnacles
Cruise ship 4	stabilizer	10 ^ 3	10	barnacles
Cutter	intake grating	10 ^ 3	10	tubeworms

Other niche areas tended to be an order of magnitude lower in terms of abundance (thousands of organisms), and this may be a function of the size of the areas in question relative to thruster and stabilizer locations. For example, dense mussel aggregations were recorded within ladder holes of barges, including approximately 531 mussels in one ladder hole of approximately 18cm diameter and 18cm depth. This mussel matt (*Barge 1*) was associated with 21 other species and over 200 barnacle individuals. The sea chest of *Barge 2* had approximately 600 organisms within it representing 14 species while thousands of organisms were estimated within the sea chest of *Cruise ship 3*. Intake gratings, dock blocks and struts (Table 3, Appendix B) were also notable hotspots of biofouling accumulation on other vessels.

Comparison with previous Containership study

In our previous study of 22 containerships (Davidson et al., 2009b), all ships were sampled in-water and sample collections (and species richness) were not possible for those sampled by remotely operated vehicle. Biofouling richness among five vessels with species-level data ranged from zero to 20 and biofouling extent was low (< 2% cover) among all 22 vessels bar one. In the present study, barges and cruise ships had greater ranges of richness; 0-33 species and 6-35 species, respectively. The important difference in sampling, especially for containerships (all in water) and cruise ships (all on dry dock) must be accounted for when comparisons are made, however. The high estimates of biofouling abundance and richness associated with niche areas of cruise ships, for example, were made from dry-dock sampling. Long inter-dry-docking-durations are likely to have played a role in this accumulation (compared to in-water sampling of containerships). Nonetheless, a general similarity among both studies was the typically unfouled surface areas of hull and concentrations of organisms around niche areas.

Discussion & Conclusions

- Transfers of species via biofouling of commercial vessels are undoubtedly an unintentional consequence of overseas and coastwise shipping on the US West Coast. We recorded at least 25 species that are non-native to the West Coast on the hulls and underwater surfaces of vessels examined. This included nine species that are not known to occur on the West Coast, so these incursion represent potential novel introductions to the coast. Movements of already established NIS may provide an opportunity for range expansions of their Pacific Coast distributions.

- Biofouling extent was generally low across the hull surfaces of vessels, but high diversity and abundance associated with heterogeneous niche areas was recorded. This is a common feature of the modern biofouling literature and challenges toward reducing biofouling accumulation at niche areas remain.

- The range of biofouling extent and richness of the vessels from the present study was greater than containerships from a previous study (Davidson et al., 2009b), albeit with some differences in sampling. We cannot conclude from this study that shorter distances or other factors related to coastwise shipping contribute to higher levels of fouling compared to ocean-going containerships, however. Previous studies have indicated that voyage distances are important determinants of biofouling accumulation and transfer because vessels traversing large expanses of sea are likely to accumulate and retain less biofouling compared to more regional vessels (Coutts & Taylor, 2004). Additional sampling (underway) of ships may allow for more robust comparisons across ship types in the future.

- The most striking aspect of biofouling recorded in this study was not related to differences among vessel types, antifouling or the effect of duration-since-dry-docking, but the influence of niche areas for harboring diverse assemblages of organisms. Even on barges that have relatively homogeneous flat surfaces, four simple ladder holes below the waterline that act as refugia from strong laminar water flow supported thousands of organisms from at least 23 different species. Similarly, while the large surface areas of cruise ship hulls we examined were largely free of macro-fouling, the areas around thrusters were colonized by tens of thousands of individuals from several different species. Antifouling paints, duration-since-dry-docking, speeds, port durations, voyage distances and geographic locations (warm vs temperate ports) undoubtedly play a role in biofouling accumulation on maritime vessels, but without heterogeneous niche areas, it is unlikely that organism transfers via biofouling would be as prevalent. The role of niche areas is consistently recorded across studies (Coutts & Taylor, 2004; Davidson et al., 2009b; Sylvester & MacIssac, 2010).

- Sampling during this study uncovered additional niche areas on vessels that we were not aware of prior to this study. To our knowledge, ladder holes below the water line of barges and large removable conical plates of azipods have not previously been highlighted as refugia for biofouling accumulation. Our sampling, albeit of just one azipod and seven barges, indicates that these niche locations provide suitable conditions for transfers of both sessile and mobile species.

- Although our sample size was small, our sampling of sea chests aligned with previous sea chest sampling in the literature (e.g. Coutts & Dodgshun, 2007). For the two vessels on which we examined sea chests, there were approximately three times the numbers of species in sea chests compared to the outer submerged surfaces of the same vessels. The diversity and abundance of organisms in these areas represents an important sub-vector and invasion risk.

- We found no relationship between biofouling and duration-since-last-dry-dock within or among vessel-types sampled. The role of antifouling paint age and its use as a predictor of species transfers by ships is elusive in the biofouling literature. While it undoubtedly plays an important role in determining biofouling extent and richness, the exact relationship between paint age and biofouling species transfers (generally or within regions, ship-types etc) has not been established. Only additional data from in-service ships will provide clues as to the shape of the relationship between paint deterioration and propagule accumulation and delivery (e.g. linear accumulation of species over time versus a tipping point).

- Biofouling vector strength is known to be relatively high compared to other maritime vectors (Hewitt & Campbell, 2009). However, there is a disconnection between retrospective analyses of biofouling invasions (measures of vector strength) and direct studies of vector activity. As the biofouling literature grows, it appears increasingly clear that niche areas are important areas of concern; the thousands of organisms at high density (close proximity) recorded from niche areas in this study and several others probably helps explain why biofouling continues as a leading cause of marine invasions. Data from niche areas may be useful for experimental approaches aimed at improving our understanding of propagule pressure and dose-response relationships in marine systems.

- At present, it appears that management strategies may best affect propagule delivery via biofouling by focusing on: a) the minority of vessels that are heavily fouled, and yet recorded in most studies with sampling of 10 or more vessels; and b) policies that target niche areas for additional maintenance attention by shippers. The stochastic highly fouled vessels operate in unusual circumstances but their biofouling extent is often manageable (i.e. steps can be taken to remove large biomass) if prior notice of vessel movements can be made. For niche area biofouling, which is a feature of most vessels, the first difficulty lies in incentivizing their cleanliness. Unlike for laminar hull surfaces, ship propulsion is not significantly reduced by niche area fouling. Therefore, niche areas are not a high priority for biofouling management between dry-dockings. The second issue is that large in-water cleaning technology exists mainly for homogeneous surfaces, and at present, little more than manual scrubbing by divers can remove fouling from most niche areas. Sea-chest protection systems offer an alternative approach that may provide a template for management other niche areas between dry-docking intervals.

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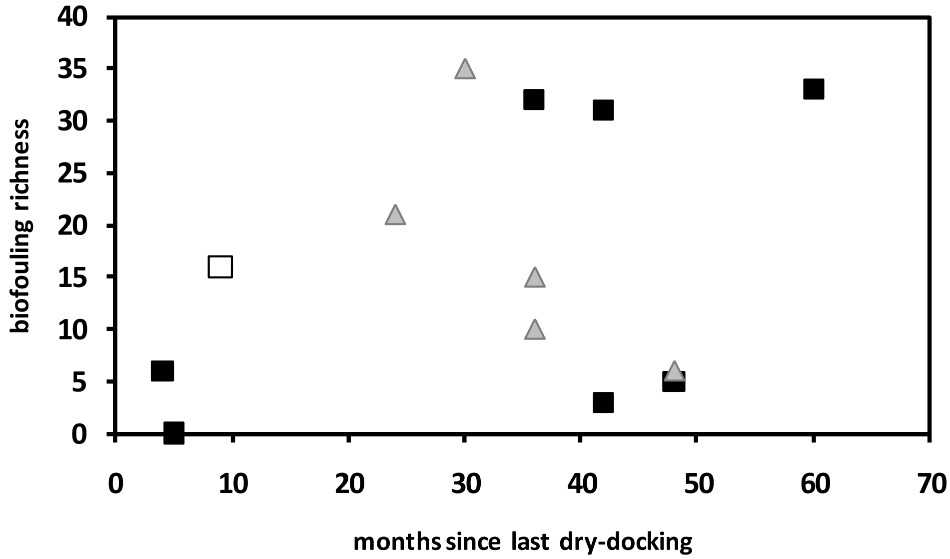
Appendix A



Plot of duration since dry-docking with richness of biofouling species per vessel. There was no significant relationship ($r^2 = 0.097$, $p > 0.05$). Barges, cruise ships and the Cutter are represented by black squares, grey triangles and a white square, respectively.

Appendix B

Species recorded from 16 vessels sampled with notes on hull locations where organisms were distributed and on non-native species established and not yet known to occur on the US West Coast.

Taxa	Hull locations where organisms occurred	US Pacific Coast Status
Algae		
Filamentous green algae	hull	
Fucus spiralis	thrusters	NIS, not established
unidentified red algae	hull, struts	
unidentified brown algae	hull, struts	
Amphipods		
Americorophium brevis	ladder holes, hull	
Corophiidae sp.	thrusters, ladder holes	
Corophium sp.	hull, sea chest	
Corophium spinicorne	ladder holes, sea chest	
Caprellid (spine?)	hull, struts	
Caprellid (no spine)	hull, struts	
Caprellid (no notch)	struts	
unidentified Caprellids	hull, dock blocks	
Desdimelita californica	ladder holes	
Desdimelita desdichada	propeller cone	
Elasmopus rapax	hull, intake grates, sea chest	NIS, established
Eogammarus confervicolus	sea chest	
Ericthonius sp.	sea chest	
Gammaridae sp.	hull, struts, sea chest, ladder holes	
Gnathopleustes pugettensis	hull	
Ischyroceridae sp.	sea chest	
Jassa marmorata	intake grate	NIS, established
Jassa staudei	ladder holes	
Monocorophium acherusicum	intake grate, sea chest, thrusters	NIS, established
Monocorophium insidiosum	sea chest	NIS, established
Photis sp.	hull	
Stenothoidae sp.	intake grate	
Anemones		
Actiniaria sp.	sea chest	
Diadumene sp.	intake grate, sea chest	
Metridium senile	ladder holes	
unidentified anemone sp1	hull	
unidentified anemone sp2	hull, thrusters	
Ascidians		
Ciona inestinalis	sea chest	NIS, established
Botrylloides cf violaceous	hull	NIS, established
Botryllus schlosseri	hull	NIS, established
unidentified solitary ascidian	hull	
Diplosoma sp	hull	

Taxa	Hull locations where organisms occurred	US Pacific Coast Status
Barnacles		
Amphibalanus amphitrite	hull, thrusters, intake grate, sea chest, propeller cone	NIS, established
Amphibalanus reticulatus	hull, thrusters, propeller cone, intake grate	NIS, not established
Balanomorpha sp1	hull, struts, sea chest, ladder holes	
Balanus crenatus	hull, sea chest, intake grate, thrusters, propeller cone, ladder holes	
Balanus perforatus	thruster	NIS, not established
Balanus trigonus	hull, intake grate, sea chest, thrusters, propeller cone	
Chthamalus dalli	hull, ladder holes	
Chthamalus fragilis	hull	NIS, not established
Chthamalus southwardum	hull	NIS, not established
Conchoderma virgatum	thrusters	
Conchoderma auritum	hull, thrusters	
Elminius modestus	thrusters	NIS, not established
Lepas anatifera	hull	
Megabalanus californicus	hull, thrusters	
Megabalanus coccopoma	hull, intake grate, sea chest, thrusters	NIS, not established
Megabalanus peninsularis	hull, intake grate	
Megabalanus sp.	hull, propeller cone, intake grate	
Pollicipes polymerus	hull, thrusters, ladder holes	
Semibalanus cariosus	hull, sea chest	
unidentified barnacle	hull	
Bivalves		
Chiona squamosa	intake grate, sea chest	
Crassostrea gigas	intake grate, sea chest, propeller cone	NIS, established
Crassostrea sp.	propeller cone	
Hiatella arctica	hull, sea chest, intake grate, propeller cone	
Isognomon sp. 1	hull sea chest	
Kellia suborbicularis	hull, intake grate, sea chest	
Limidae sp. 1	propeller cone	
Lithophaga sp.	intake grate, sea chest	
Lopha sp. 1	hull	
Mytilus sp.	hull, intake grate, sea chest, thrusters, propeller cone, ladder holes	
Ostrea edulis	propeller cone	NIS, established
Ostreidae sp.	sea chest, propeller cone	
Ostreidae sp. 1	hull	
Pinnidae sp.	intake grate, sea chest	
Pteria sp.	hull, intake grate, sea chest	
Pteria sterna	hull, intake grate, sea chest	status?
Septifer sp.	sea chest	
Sphenia sp.	sea chest	
unidentified bivalve *(Gemma sp?*	thrusters	

Taxa	Hull locations where organisms occurred	US Pacific Coast Status
Bryozoans		
Aetea sp	hull, struts	
Alcyonidium sp.	ladder holes	
Bowerbankia gracilis	hull, sea chest	
unidentified bryozoan	sea chest	
Bugula sp.	hull, sea chest	
Bugula neritina	hull	NIS, established
Caulibugula occidentalis	thrusters	
Celleporaria sp.	hull	
Celleporella sp?	hull	
Crisulipora sp?	hull	
Conopeum sp	hull, ladder holes	
Conopeum cf. reticulum	ladder holes	
Corynoporella sp?	hull	
Cryptosula cf pallasiana	hull	NIS, established
Fenestrulina sp	hull	
Lagenicella sp.	sea chest	
Lichenopora sp.	hull, sea chest	
Membranipora sp.	hull, struts, thrusters, sea chest	
Schizoporella japonica	hull	NIS, established
Schizoporella sp (pugens?)	hull	
Scrupocellaria sp.	strut, thruster	
Tegella armifera	sea chest	
Tricellaria occidentalis	propeller cone	
Watersipora sp	hull, struts	
Watersipora subtorquata	thrusters	NIS, established
unidentified encrusting bryo A	hull, struts	
unidentified encrusting bryo B	hull	
unidentified encrusting bryo C	hull, struts	
unidentified encrusting bryo D	dock block areas	
unidentified arborescent bryo	hull	
Mobile crustaceans		
Brachyura sp.	sea chest, thrusters	
Calanus marshallae	intake grate	
Crustacea sp.	hull	
Euphausia pacifica	intake grate, sea chest	
Gnorimosphaeroma oregonense	hull, sea chest, ladder holes	
Gnorimosphaeroma sp.	hull	
Neocalanus cristatus	intake grate, sea chest	
Neocalanus plumchrus	sea chest	
Pasiphaea pacifica	propeller cone	
Plagusia sp.	thrusters	
Sphaeromatid sp	hull, ladder holes	
Tanaid spp	hull, strut, ladder holes, dock blocks	
Gastropods		
Crepidula aculeata	sea chest	
Lottia digitalis	hull	
Stramonita sp?	thrusters	
Velutina sp?	thrusers	
unidentified Nudibranch	hull	

Taxa	Hull locations where organisms occurred	US Pacific Coast Status
Hydroids		
Eudendrium californicum	thrusters	
Hydrozoa spp.	hull, struts, ladder holes, sea chest, intake grate, dock blocks	
Hydroida sp.	thrusters	
Obelia dichotoma	hull, sea chest, intake grate, propeller cone, ladder hole	
Obelia sp.	ladder hole	
Tubularia sp?	hull	
Insects		
Coleoptera sp.	sea chest	
Cyclopodia sp.	sea chest	
Diptera sp.	hull	
Paraclunio alaskensis	hull, ladder hole	
Nematodes		
Nematoda sp.	hull	
Nemerteans		
Emplectonema gracile	hull, ladder holes	
Nemertea sp.	hull	
unidentified Nemertean spp	hull, struts	
Platyhelminthes		
unidentified flatworm (white)	hull	
unidentified flatworm (brown)	struts	
unidentified flatworm	hull	
Polychaetes		
Autolytus sp.	hull	
Capitella capitata complex	sea chest	
Caulleriella pacifica	sea chest	
Dorvilleidae sp.	sea chest	
Ficopomatus sp	hull	NIS, likely established
Hydroides elegans	intake grate	NIS, established
Lepidonotus sp.	sea chest	
Lepidonotus squamatus	intake grate, sea chest	
Micropodarke dubia	intake grate, sea chest	
Naineris dendritica	intake grate, sea chest	
Nereidae spp.	hull, struts, ladder holes, intake grate, dock blocks	
Nereis vexillosa	ladder holes	
Paleanotus bellis	hull, lader holes	
Phyllochaetopterus sp.	sea chest	
Phyllodoce williamsi	ladder holes	
Polychaete sp.	sea chest	
Polydora sp.	sea chest	
Polynoidae sp.	sea chest	
Pomatoleios sp.	sea chest	
Proceraea sp.	hull	
Sabellid sp	hull	
Serpulidae sp.	thrusters, propeller cone, sea chest	
Spirorbid sp.	thrusters	
Typosyllis adamanteus	hull	
Pycnogonid		
Ammothea hilgendorfi	sea chest	
Ammothella pacifica	sea chest	NIS, not established
Endeis spinosa	sea chest	NIS, not established
Porifera		
unidentified sponge	thrusters	
Unidentified larval form	sea chest	